37 °C with gentle tapping,

3) mixing with an equal volume of Tris-HCl and EDTA buffer saturated phenol,

4) centrifuging at approximately 3,000 rpm for approximately 10 min, and collecting the upper aquous phase containing DNA,

5) mixing with equal volume of phenol and chloroform,

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6) centrifuging at approximately 3,000 rpm for approximately 10 min, and collecting the upper aquous phase containing DNA,

7) mixing with an equal volume of chloroform/isoamyl alcohol prepared at a ratio of 96:4,

8) centrifuging at approximately 3,000 rpm for approximately 10 min,

9) collecting the upper aquous phase containing DNA and precipitating using a salt and ethanol,

10) cenrifuging at approximately 13,000 rpm for approximately 30 min,

11) removing the upper liquid phase and air drying the bottom DNA pellet,

12) dissolving the air dried DNA pellets in Tris-HCl and EDTA buffer.

4. (Amended) The method of Claim 9 wherein the method further includes the steps of analyzing the biologically active transcription factors present in the top clear supernatant of step *I*.

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Insert Claim 9 (Newly submitted). Claim 9 which is the same as the **Examiner's proposed Amendment of Claim 2** (previously submitted) except for minor changes as noted in the REMARKS.

9. (Newly submitted) A method for simultaneous isolation of biologically active transcription factors and DNA, wherein no ultracentrifugation or sonication is used, comprising the following steps:

a) collect cells from a culture or from a patient,

b) washing said cells at least once with PBS,

c) suspend and maintain said cells in buffer A (cell lysis buffer) for approximately 15 minutes wherein buffer A comprises: 20 mM Hepes, pH 7.9, 10 mM NaCl, 3 mM $MgCl_2$, 0.1% NP-40, 10% glycerol, 0.2 mM EDTA, 1 mM DTT, 0.4 mM PMSF, 1 μg/ml antipain, 1 μg/ml leupeptin,

d) centrifuge the suspension of step c at approximately 2,000 rpm for approximately 5 min at approximately 4 °C,

e) remove the upper cytoplasmic supernatant fraction and then clarify this fraction by adding buffer D (cytoplasmic extraction clarification buffer) at approximately 4 °C, wherein buffer D comprises: 20 mM Hepes, pH 7.9, 400 mM NaCl, 0.2 mM EDTA, 40% glycerol, 1 mM DTT, 0.4 mM PMSF, 1 μg/ml antipain , 1 μg/ml leupeptin,

f) centrifuge the clarified fraction formed in step e approximately 13,000 rpm for approximately 15 min,

g) remove and freeze the top clear supernatant on dry ice then store the frozen top clear supernatant at approximately -86 °C,

h) wash the bottom nuclear fraction formed at the end of step d with buffer B (extraction buffer without salt) wherein buffer B comprises: 20 mM Hepes, pH 7.9, 0.2 mM EDTA, 20% glycerol, 1 mM DTT, 0.4 mM PMSF, 1 μg/ml antipain , 1 μg/ml leupeptin,

i) centrifuge the mixture formed in step h at approximately 2,000 rpm for approximately 5

minutes at approximately 4 °C, thereby pelleting cellular nuclei,

j) suspend the pelleted nuclei of step i in buffer C (extraction buffer with salt) on ice, tapping the suspended mixture for approximately 45 minutes whereby nuclear proteins are extracted, wherein buffer C comprises: 20 mM Hepes, pH 7.9, 400 mM NaCl, 0.2 mM EDTA, 20% glycerol, 1 mM DTT, 0.4 mM PMSF, 1 μg/ml antipain , 1 μg/ml leupeptin,

k) centrifuge the mixture formed in step j at approximately 13,000 rpm for approximately 15 minutes, at approximately 4 °C,

l) remove the top clear supernatant comprising biologically active transcription factors from the bottom nuclear fraction containing nucleic acids, and quick freeze on dry ice and store at approximately -86°C,

m) extract DNA from said bottom nuclear fraction.

Insert Claims 10 and 11 (Newly submitted).

10. (Newly submitted) The method of Claim 9 wherein the method further includes: the steps of analyzing the biologically active cytoplasmic factors present in the top clear supernatant of step g.

11. (Newly submitted) The method of Claim 9 wherein the step *l*, as proposed by the Examiner, is followed by the step of: , and quick freeze on dry ice and store at approximately -86°C.